REMARKS

After entry of this amendment, claims 1, and 3-63 will be pending in this application. Claims 19-63 have been withdrawn from consideration as drawn to a non-elected invention. The specification has been amended to consistently recite the same reference numerals in relation to the same features shown in the drawings. Claim 1 has been amended to recite the elements of claim 2, which has been cancelled.

Because claim 1 now covers the exact subject matter of previous claim 2, the amendments do not narrow the scope of previous claim 2. Accordingly, scope of the invention as previously claimed in claim 2 is not limited and the invention is entitled to the same, or broader, scope either literally or under the Doctrine of Equivalents.

Amendment of the specification, claims and drawings is supported by the application as filed, does not add new matter, and is otherwise proper. Applicants respectfully request entry of this amendment in its entirety.

In view of the amendment and following remarks, Applicants respectfully request reconsideration of the application and claims and submit that the application is in condition for allowance.

I. Objection to the Drawings

In the Office Action, the drawings were objected to because FIG. 3 "include[d] the following reference sign(s) not mentioned in the description: ref. #21 if fig. 3." Applicants respectfully disagree with the objection and would like to draw the Examiner's attention to page 13, lines 5-6 of the specification as originally filed which recites reference numeral 21 in the following passage: "The substrate 20 is formed of a base plate of material 21..." which has been amended to read, "The substrate 21 is formed of a base plate of material, e.g., a polymeric plastic...." Applicants further note that in paragraph 5 of the Office Action, the Examiner noted that reference character 21 (Pg. 16, line 28-29) was used to refer to a substrate.

The Office Action also objected to FIG. 3 because it did "not include the following reference sign(s) mentioned in the description: ref. #33 of fig.3." Applicants include herewith a proposed drawing change showing reference numeral 33 in red. A clean copy of amended FIG. 3 is also included with this Amendment along with a Letter Regarding Proposed Changes to Drawings.

The drawings were further objected to "because reference characters '20' (pg. 13, line 5) and '21' (pg. 16, line 28-29) have both been used to designate a substrate." The specification has been amended to always recite reference numeral 21 when referring to a substrate.

Finally, FIG. 3 was objected to "because reference character '33' has been used to designate both a layer (pg. 17, line 25-26) and a film (page 17, line 31). The specification has been amended to recite only a layer when referring to reference numeral 33.

Applicants submit that the proposed drawing changes and amendments to the specification fully meet the objections to the drawings and respectfully request the Examiner withdraw these objections.

II. Objection to the Claims

In the Office Action, claim 4 was objected to as being improperly dependent on itself. Claim 4 has been amended to depend from claim 3 and Applicants respectfully request that the Examiner withdraw this objection.

III. Claim Rejections Under 35 U.S.C. §112

Claims 1-18 were "rejected under 35 U.S.C. 112, second paragraph, as being indefinite... in that there is no method steps in the detection of the pathogen [recited in claim 1.]" Claim 2 was not rejected under the second paragraph of 35 U.S.C. § 112. Claim 1 has been amended to incorporate the method limitations previously recited in claim 2 including,

"(c) applying a sample to be tested for the presence of the specific pathogen to the surface of the detection region of the substrate; and

(d) thereafter applying the liquid crystal material to the detection region that will be aligned by the microstructures on the surface of the substrate in the absence of binding of pathogen particles to the surface of the substrate, whereby the presence of the selected pathogen in the sample will be manifested by a visually observable disordering of the liquid crystal material caused by the pathogen particles bound to the substrate." Applicants respectfully submit that the amendment of claim 1 overcomes the rejection set forth in Office Action.

Claim 1 was also rejected because "it is unclear the relationship that the liquid crystal has with the substrate. Is it a bi-layer that attaches to the substrate or a covering for the substrate?" As stated above, claim 1 has been amended to recite the method limitations previously found in claim 2 which was not rejected under the second paragraph of 35 U.S.C. § 112 and only requires that a liquid crystal be applied to the detection region. The interaction of the liquid crystal and the substrate is not claimed, nor is it crucial for the present invention other than the requirement that "the presence of the selected pathogen in the sample will be manifested by a visually observable disordering of the liquid crystal material caused by the pathogen particles bound to the substrate." Applicants respectfully submit that the amendment of claim 1 overcomes the rejection of this claim under the second paragraph of 35 U.S.C. § 112.

Claims 3-7, 10, 15, and 11-13 were also rejected in the Office Action. First, claims 3-7 were rejected because "it is unclear what the method steps are for the detection of the pathogen of claim 3-7 since it is just claiming the type of substrate." Second, claims 10 and 15 were rejected because these claims "are also claiming the size of the depression but are unclear on how this correlates with the method step of detecting the pathogen. Since such a claim relates to the device and not the method of detection" Finally, claims 11-13 were rejected as "just claiming the size of the grooves." Therefore, the Office Action seems to indicate that these claims were each rejected because they further define a device or apparatus used in the method such as the substrate (claims 3-7), the size of the depression (claims 10 and 15), or the size of the grooves (claims 11-13). Applicants first note that the method of amended claim 1 is drawn to a method for use in detecting the presence of a selected

microscopic pathogen. As with all embodied methods, devices or apparatuses are used in the method. For example, amended method claim 1 includes, "providing a substrate having a detection region thereon...," "treating the surface of the detection region to provide a layer thereon that blocks non-specific binding...," "applying a sample to be tested for the presence of the specific pathogen to the surface of the detection region of the substrate," and "thereafter applying the liquid crystal material to the detection region that will be aligned by the microstructures on the surface of the substrate in the absence of binding of pathogen particles to the surface of the substrate,...." Rejected dependent claims 3-7, 10, 11-13, and 15 further define the method of claim 1 because they further describe the devices used in the method. Therefore, each of the rejected dependent claims properly narrows the scope of amended method claim 1. Applicants respectfully note that this is entirely proper procedure for dependent claim practice and that these dependent claims reasonably apprise one of skill in the art of the scope of the invention. For this reason, Applicants respectfully contend that the rejection of these claims is improper under the second paragraph of 35 U.S.C. § 112.

With respect to the above comments, Applicants first note that claim 1, from which claims 3-7, 10, 11-13, and 15 each depend, has been amended to recite the additional method limitations of claim 2 related to "applying a sample..." and "thereafter applying the liquid crystal material...." Claim 3 depends from amended claim 1 and further specifies, "coating at least a portion of the detection region with an inorganic material selected from" a recited group of inorganic materials. Amended claim 4 depends from claim 3 and further specifies, "the inorganic material is silver or gold and the method further comprises treating at least a portion of the silver or gold with a mercaptan or a disulfide." Claim 5 depends from amended claim 1 and further specifies, "the substrate is formed of a molded polymer plastic." Claim 6 depends from claim 5 and further specifies, "the molded polymer plastic comprises polystyrene, polycyanoacrylate, or polyurethane." Claim 7 depends from claim 5 and further specifies, "the molded polymer is polydimethylsiloxane." Claim 10 depends from amended claim 1 further specifying, "the selected pathogen is a virus and the depressions on the surface of the detection region have a width and depth in the range of 5 nm to 500 nm." Claim 11 depends from claim 1 further specifying, "the depression on the surface of the detection region

of the substrate comprise parallel grooves having a width of approximately 100 nm." Claim 12 depends from claim 11 further specifying, "the grooves are separated by ridges having a width of about 100 nm." Claim 13 depends from claim 11 further specifying, "the grooves have a depth of approximately 100 nm." Finally, claim 15 depends from claim 1 and further species, "the selected pathogen is a bacteria and the depressions on the surface of the detection region have a width and depth in the range of 0.1 μm to 10 μm."

One skilled in the art would recognize that the size of the depressions are chosen "to be occupied by the selected pathogen" based on the size of the selected pathogen, and that viruses generally range in size from 5 to 500 nm and bacteria generally range in dimension from 0.1 μm to 10 μm. Thus, where a specific pathogen is to be detected, the depression dimensions of the substrate would generally be chosen based on the dimensions of that specific pathogen so that the pathogens, when present, can at least partially reside in the depressions.

Accordingly, because the claims "apprise[] one of ordinary skill in the art of [their] scope and, therefore, serve the notice function required by 35 U.S.C. §112, second paragraph" (MPEP §2173.02), the Examiner is respectfully requested to withdraw these rejections.

IV. Rejection of Claims Under 35 U.S.C. §102

In the Office Action, claims 1-7, 14 and 16 were "rejected under 35 U.S.C. 102(e) as being anticipated by Abbott *et al.* (U.S. Patent 6,284,197 B1)" because:

> Abbot *et al.* anticipated the claimed invention by teaching a method for detection of an analyte. The method steps include providing a substrate (a first substrate), a binding agent (a recognition moiety), and the detection of the analyte is due to the interaction of the analyte with the binding agent on the substrate. This binding causes a visual distortion of the liquid crystal (mesogens) enabling the detection of the analyte (col. 14, lines 16-43). The analyte is a biomolecule (pathogen) (col. 26, lines 21-23; col. 29, lines 11-15). The detection region (substrate) is coated with gold (gold film) and treated with disulfide (col. 22, lines 4-39). The substrate is formed from a polymer, preferably polydimethylsiloxane (col. 15, lines 66-67 and continue to col. 16, lines 1-14). The binding agent is a peptide, antibody, or protein (col. 26, lines 30-36; col. 28, lines 53-67). The binding agents are located in the depression of the detection region (col. 40, lines 42-44).

According to the MPEP a "claim is anticipated only if each and every element as set forth in the claim is found... in a single prior art reference. The **identical invention must** be shown **in as complete detail** as is contained in the... claim." MPEP § 2131 (emphasis added)(citations omitted). Specifically, Abbot *et al.* fail to teach that their detection system is suitable for detecting a pathogen, i.e., "[a]ny microorganism which, by direct interaction with (infection of) another organism (by convention a multicellular organism) causes disease in that organism." SINGLETON, P., *et al.*, DICTIONARY OF MICROBIOLOGY AND MOLECULAR BIOLOGY 641 (1991)(a copy of which is enclosed for the Examiner's convenience). Abbott *et al.* do state at column 42, lines 40-49:

> [w]hen the analyte is a biomolecule, any recognition moiety which interacts with the biomolecule is useful in practicing the present invention. Thus, when the analyte is a nucleic acid, in one embodiment, the recognition moiety is a nucleic acid having a sequence which is at least partially complementary to the recognition moiety sequence. When the recognition moiety is a peptide, an antibody specific for that peptide can be used as the analyte. In another preferred embodiment, a protein, other than an antibody (e.g., enzyme, receptor) is the analyte.

However, Abbott *et al.* fail to disclose any biomolecule which is a pathogen or indeed any method for detecting the presence of a pathogen in a sample. Furthermore, the cited reference fails to teach, suggest, or disclose any method in which the presence of a selected pathogen in a sample will be manifested by a visually observable disordering of the liquid crystal material caused by the pathogen particles bound to the substrate. Moreover, all the biomolecules recited by Abbott *et al.* are small and non-complex compared to a pathogen. Because Abbott *et al.* fail to teach that a pathogen can be detected, Abbott *et al.* must also fail to teach a substrate including depressions of suitable size to occupied by the selected pathogen. Furthermore, the cited reference simply fails to disclose any mechanism or method for detecting pathogens such as viruses or bacteria.

Claims 1-6 and 14 were also "rejected under 35 U.S.C. 102(e) as being anticipated by Abbott *et al.* (US Patent 6,277,489 B1)" because:

> Abbot *et al.* anticipated the claimed invention by teaching a method for detection of an analyte. The method steps include providing a substrate (a first substrate), a binding agent (a recognition moiety), and the detection of the analyte is due to the interaction of the analyte with the binding agent on the substrate. This

> binding causes a visual distortion of the liquid crystal (mesogens) enabling the detection of the analyte (col. 14, lines 16-43). The analyte is a biomolecule (pathogen) (col. 51, lines 27-31). The detection region (substrate) is coated with gold (gold film) and treated with disulfide (col. 11, lines 34-37 and 51-55). The substrate is formed from a polymer (col. 11, lines 8-18). The binding agent is a peptide or antibody (col. 19, lines 56-59; col. 26, lines 18-25).

As noted above, Abbott *et al.* fail to teach any analyte that is a pathogen. Therefore, as described above, with respect to U.S. Patent No. 6,284,197 B1, this reference fails to teach, disclose, or suggest any method for detecting pathogens or any method by which the presence of a selected pathogen in a sample is manifested by a visually observable disordering of a liquid crystal material caused by the pathogen particles bound to the substrate. For these reasons, the rejection must fail.

Claims 1-17 and 14 were also "rejected under 35 U.S.C. 102(f)" over U.S. Patent No. 6,284,197. However, as discussed above, U.S. Patent No. 6,284,197 does not disclose each and every element of the claimed invention and cannot serve as a proper anticipatory reference. For this reason, the rejection must fail.

Because both Abbott *et al.* references fail to teach the "identical invention... in as complete detail as is contained in the... claim" they cannot anticipate the present claims and Applicants respectfully request that the Examiner withdraw the rejection of these claims.

V. Rejection of Claims Under 35 U.S.C. §103

In the Office Action, claims 8-9 were "rejected under 35 U.S.C. 103(a) as being obvious over Abbott *et al.* (US Patent 6,284,197 B1) in view of Leavitt *et al.* (US Patent 5,712,103) because "Leavitt *et al.* teaches a method step where an assay involves and antigen or antibody (immunoglobulin) immobilized on a substrate and to minimize non-specific binding by coating the substrate with bovine serum albumin."

As stated in the MPEP "[t]o establish *prima facie* obviousness of a claimed invention, all the claim limitations must be taught or suggested by the prior art." MPEP § 2143.03. However, as discussed above Abbott *et al.* fail to teach or suggest all the elements of

the claimed invention, specifically that the biomolecule is a pathogen. Leavitt *et al.* fail to make up for this deficiency as they do not teach any method for detecting a pathogen.

Claims 10-13 and 15 were "rejected under 35 U.S.C. 103(a) as being obvious over Abbott *et al.* (US Patent 6,284,197 B1)." However, as discussed above Abbott *et al.* fail to teach or suggest all the elements of the claimed invention, specifically that the biomolecule is a pathogen. Likewise, Abbott *et al.* alone cannot teach or suggest having the claimed depression sizes, which correspond to sizes capable of at least partially holding pathogens, because Abbott *et al.* fail to teach or suggest any depression which purpose is to be occupied by the selected pathogen or a method for detecting a pathogen.

Claims 17-18 were "rejected under 35 U.S.C. 103(a) as being unpatentable over Abbott *et al.* (US Patent 6,284,197 B1) in view of Leavitt *et al.* (US Patent No. 5,712,103) as applied to claims 8-9 above, and further in view of Chagnon *et al.* (US Patent No. 4,628,037) because "[t]he method of Abbott *et al.* as modified by Leavitt *et al.* differs from the claimed invention in failing to specifically include the use of magnetic beads. Chagnon *et al.* teaches a binding assay method that involves using magnetic particles (beads) to measure the amount of analyte (col. 9, lines 58-64)." However, Abbott *et al.* in combination with Leavitt *et al.* do not teach or suggest all the claim elements, such as detection of a pathogen. Chagnon *et al.* do not overcome these deficiencies merely by teaching the use of magnetic particles. The combined references simply fail to teach or suggest any method for detecting pathogens using a substrate in combination with magnetic beads and liquid crystals or indeed providing any reason why one would want to so or expect success in such an undisclosed endeavor. "The mere fact that references can be combined or modified does not render the resultant combination obvious unless the prior art suggests the desirability of the combination." MPEP § 2143.01. There is no teaching or suggestion in any of the references to combine them and achieve the claimed invention. Additionally, none of the references teach or suggest contacting the beads with a substrate or providing depressions of a size sufficient to be occupied by the magnetic beads.

Accordingly, the references cited in the Office Action fail to teach or suggest all of the claim elements of the present invention and Applicants respectfully request the Examiner withdraw these rejections.

VI. Double Patenting Rejection

In the Office Action, claims 1 and 14 were "rejected under the judicially created doctrine of obviousness-type double patenting as being unpatentable over claims 1 and 5-7 of U.S. Patent 6,284,197 (Abbott *et al.*)." Applicants first note that claim 1 has been amended to include each of the limitations of now cancelled claim 2. The amendment of claim 1 should thus overcome the rejection of claim 1 and claim 14 which depends from amended claim 1 and which, therefore, contains each and every limitation of amended claim 1. Applicants respectfully disagree with this rejection and submit that there are several points of non-obviousness between the present claims and claims 1 and 5-7 of Abbott *et al.* First, the cited claims of Abbott *et al.* require "said recognition moiety bound to said self-assembled monolayer" which the present claims do not. Second the present claims include depressions "of a size sufficient to be occupied by the selected pathogen", a layer that provides "non-specific binding" and detection of a selected pathogen. Therefore, the claims of Abbott *et al.* neither teach nor suggest the features set forth in amended claim 1 and claim 14. Thus, Applicants respectfully submit the claims of the present invention are not obvious of claims 1 and 5-7 of Abbott *et al.*

CONCLUSION

In view of the above remarks and amendments, it is respectfully submitted that this application is in condition for allowance. Early notice to that effect is earnestly solicited. The Examiner is invited to telephone the undersigned at the number listed below if the Examiner believes such would be helpful in advancing the application to issue.

Respectfully submitted,

Date July 3, 2002

By _____

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Version with markings to show changes made

In the specification:

Paragraph starting on page 13, line 3:

For purposes of exemplifying the invention, a simplified plan view of the substrate of a detection apparatus in accordance with the invention is shown generally at 20 in FIG. 1. The substrate **[20]** 21 is formed of a base plate of material **[21]**, e.g., a polymeric plastic, preferably having a flat top surface area 22 which surrounds a detection region 23. As discussed further below, the detection region 23 is formed of a microstructure having depressions, which as shown in FIGS. 1 and 2, may comprise adjacent ridges 25 separating depressions, preferably grooves 26, with the grooves disposed parallel to one another across the detection region 23. The width and depth of the grooves 26, and the spacing between the grooves as defined by the tops of the ridges 25, are selected to cause liquid crystal material to adopt a uniform orientation that is impressed upon it by the grooves and ridges. The size of the grooves and the spacing of the grooves are also selected such that adherence of a pathogen particle or clumps of particles of appropriate size will disrupt the uniform orientation of the liquid crystal material, causing a visible change in the appearance of a liquid crystal to signal the detection of the virus to an observer. Various conventional liquid crystal materials may be utilized, including nematic and smectic liquid crystal. The liquid crystals may be thermotropic or lyotropic phases. In general, groove widths and depths which are suitable to be occupied by viruses will be in the range of 5 to 500 nanometers (nm) and suitable spacing of the grooves 26 by the ridges 25 may also be in the same range. Where the selected pathogen is a bacteria, the width and depths of the grooves will generally be in the range of 0.1 micrometer (μm) to 10 μm to allow the grooves to be occupied by the bacteria. The grooves may be of various geometries, e.g., square, rectangular, triangular, or semicircular, and typically will be formed somewhat rounded or wedge shaped at nano-dimensions. The width of the grooves is preferably selected to be about the size of an individual pathogen particle, so that the particle will fit at least partially into a groove to occupy the groove. The depressions 26 may be formed in geometries other than linear, parallel grooves, e.g., as ellipsoids, truncated grooves,

mixtures of grooves of different width, depth and shape, selected to act on the liquid crystal to orient it in the desired uniform orientation.

Paragraph starting on page 14, line 3:

The apparatus of the present invention may utilize the grooved substrate by itself in the form 20 shown in FIG. 1. The apparatus of the invention may also be utilized with a cover panel to cover the liquid crystal material, as further illustrated in the cross-sectional view of FIG. 2. The substrate **[20] 21** is formed in the same manner as described above, having the grooves 26 spaced by ridges 25 of selected and controlled dimensions. A spacer or gasket 30 is mounted on the non-textured surface 22 to fully or partially surround the detection region 23 and to support a cover plate 31. The substrate **[20] 21**, spacer 30 and cover plate 31 enclose a volume 32 which can contain the liquid crystal material.

Paragraph starting on page 16, line 22:

For purposes of exemplifying the invention, a simplified plan view of the substrate of a detection apparatus in accordance with the invention is shown generally at 20 in FIG. 1. The substrate **[20] 21** is formed of a base plate of material **[21]**, e.g., a polymeric plastic, preferably having a flat top surface area 22 which surrounds a detection region 23. As discussed further below, the detection region 23 is formed of a microstructure having depressions, which as shown in FIGS. 1 and 2, may comprise adjacent ridges 25 separating depressions, preferably grooves 26, with the grooves disposed parallel to one another across the detection region 23. The width and depth of the grooves 26, and the spacing between the grooves as defined by the tops of the ridges 25, are selected to cause liquid crystal material to adopt a uniform orientation that is impressed upon it by the grooves and ridges. The size of the grooves and the spacing of the grooves are also selected such that adherence of a pathogen particle or clumps of particles of appropriate size will disrupt the uniform orientation of the liquid crystal material, causing a visible change in the appearance of a liquid crystal to signal the detection of the virus to an observer. Various conventional liquid crystal materials may be utilized, including nematic and smectic liquid crystal. The liquid crystals may be thermotropic or lyotropic phases. In general, groove widths and depths which are suitable to be occupied by viruses will be in the range of 5 to 500 nanometers (nm) and suitable spacing of the grooves 26

by the ridges 25 may also be in the same range. Where the selected pathogen is a bacteria, the width and depths of the grooves will generally be in the range of 0.1 micrometer (μm) to 10 μm to allow the grooves to be occupied by the bacteria. The grooves may be of various geometries, e.g., square, rectangular, triangular, or semicircular, and typically will be formed somewhat rounded or wedge shaped at nano-dimensions. The width of the grooves is preferably selected to be about the size of an individual pathogen particle, so that the particle will fit at least partially into a groove to occupy the groove. The depressions 26 may be formed in geometries other than linear, parallel grooves, e.g., as ellipsoids, truncated grooves, mixtures of grooves of different width, depth and shape, selected to act on the liquid crystal to orient it in the desired uniform orientation.

Paragraph starting on page 17, line 17:

An example of the invention will now be described with reference to FIGS. 3-8. This example utilized an elastomeric substrate **[20]** 21 formed by nanoscale molding of polydimethylsiloxane, using as a mold the surface of a silicon wafer that was patterned with grooves having approximately 100 nm width and depth and 100 nm spacing that were formed by using e-beam lithography. The resulting microstructure with substantially square grooves and ridges of 100 nm dimensions is illustrated in the simplified cross-sectional view of FIG. 3. Atomic force microscopy and scanning electron microscopy confirmed that the nanometer-scale topography of the silicon template was reproduced into the polymeric material. A layer 33 of BSA (as a blocking layer) and immunoglobulin G (IgG) (as a binding agent) was formed in a film on the surfaces of the grooves 26 and the ridges 25 in the detection region. One skilled in the art will recognize that portions or fragments of immunoglobulins may be used in place of the whole immunoglobulin. As shown in FIG. 3 for illustration, the size of the BSA and IgG molecules adhered to the surfaces to form the **[film]** layer 33 was small in comparison to the dimensions of the grooves 26. The dimensions of the grooves 26 were comparable in size to vesicular stomatitis virus (VSV) (typically virus particle size about 100 nm x 45 nm), a particle of which is shown for illustration at 35 in FIG. 3. To form the **[film]** layer 33, the patterned substrate **[20]** 21 is sequentially immersed first in an aqueous solution of IgG and then in an aqueous solution of BSA. The resulting layer of molecules on the substrate surface is

schematically illustrated in FIG. 5. When the substrate **[20]** 21 with the **[film]** layer 33 formed in this manner had a layer of liquid crystal material placed on it, the detection region in which the nanometer-scale grooves were formed caused the liquid crystal to appear uniformly dark when viewed between cross-polarizing sheets (with the grooves parallel to the analyzer). A commercially available liquid crystal material was utilized, 4-cyano-4'-pentylbiphenyl (5CB) nematic liquid crystal manufactured by BDH and available from EM Industries, Hawthorne, New York. (Other liquid crystal materials may be used, including smectic liquid crystals such as 8CB.) FIG. 4 shows the nano-textured detection region 23, the flat untextured area 24, and the region of the spacer 30. A photograph through the analyzer showed that the detection region 23 appeared uniformly dark due to uniform anchoring of the liquid crystal whereas the flat regions 22 of the substrate surface that do not have grooves were brightly colored because there are no grooves and the liquid crystal is not uniformly anchored on the surface. Thus, the presence of the nanoscale grooves 26 in the surface of the substrate in the detection region cause the liquid crystal to adopt a uniform orientation which is not erased by the adsorption of the BSA and IgG onto the substrate surface in the layer 33.

Paragraph starting on page 18, line 23:

FIGS. 5-7 schematically illustrate the results of an example utilizing such a substrate in combination with liquid crystals to detect the presence of a specific strain of virus, the Indiana strain of VSV. The surfaces of the substrate **[20]** 21 with the grooves 26 formed therein was pretreated as described above to form a layer 33 of BSA and IgG, the IgG selected to specifically bind to the Indiana strain of VSV (i.e., anti-VSV-I IgG), as illustrated schematically in FIG. 5. The width of the grooves 26 in the substrate was 100 nm, which is on the order of the size of the VSV virus particle (about 100 nm x 45 nm), allowing the virus particle to at least partially fit into and occupy the groove. A photograph and observation of the detection region only which has the liquid crystal 5CB in contact therewith, showed a uniform dark appearance indicating the uniform anchoring of the liquid crystal to the substrate surface in the detection region. Another substrate similarly pretreated with BSA and anti-VSV-I IgG then was treated by placing a droplet of buffer containing the Indiana strain of VSV onto the surface of the substrate. The droplet was confined between a glass cover slip and the

surface of the substrate. The cover slip was then removed, the surface of the substrate was rinsed with phosphate buffered saline (PBS), then placed under a stream of nitrogen to displace excess PBS, and then contacted with 5CB liquid crystal. When the droplet of buffer contained about 106 pfu/ml of VSV-I, the VSV-I was bound to the IgG, as illustrated schematically in FIG. 6, and the liquid crystal in contact with the surface then appeared non-uniform and bright. This result indicates that the presence of the virus erases the effect of the nanoscale grooves 26 on the alignment of the liquid crystal. A further experiment was performed utilizing the New Jersey strain of VSV, which does not bind to the IgG that is specific to the Indiana strain, as illustrated schematically in FIG. 7. When the substrate treated in this manner had liquid crystal applied to it, the uniform orientation of the liquid crystal was not substantially altered. The above results demonstrate that the detection system may be used to identify the presence of the Indiana strain of VSV in a sample and that the detection apparatus can differentiate between different strains of the same virus. Thus, the assay is specific to the Indiana strain of VSV.

In the claims:

1. (Amended) A method for use in detecting the presence of a selected microscopic pathogen in a sample comprising:

(a) providing a substrate having a detection region thereon comprising a surface comprising microstructures including depressions of width and depth sized to align a liquid crystal material in contact therewith and wherein the depressions are of a size sufficient to be occupied by the selected pathogen; **[and]**

(b) treating the surface of the detection region to provide a layer thereon that blocks non-specific binding of pathogens to the surface and that includes a binding agent that specifically binds the selected pathogen to be detected;

(c) applying a sample to be tested for the presence of the specific pathogen to the surface of the detection regi n of the substrate; and

(d) thereafter applying the liquid crystal material to the detection regi n that will be aligned by the microstructures n the surface f the substrate in the absence

of binding of pathogen particles to the surface of the substrate, whereby the presence of the selected pathogen in the sample will be manifested by a visually observable disordering of the liquid crystal material caused by the pathogen particles bound to the substrate.

4. (Amended) The method of Claim **[4]** **3**, wherein the inorganic material is silver or gold and the method further comprises treating at least a portion of the silver or gold with a mercaptan or a disulfide.